

We claim:

1. A system for monitoring in real time the progress of a nucleic acid amplification reaction, the system comprising:

5 a reaction mixture for carrying out the nucleic acid amplification reaction so that an amplification product is synthesized;

a sample interface comprising a fiber optic co-axially disposed with a lens so that an excitation beam transmitted by the fiber optic is focused into a volume of the reaction mixture by the lens;

10 a first fluorescent indicator capable of generating a first fluorescent signal whose intensity is proportional to the amount of an amplification product in the volume of the reaction mixture illuminated by the excitation beam; and

a second fluorescent indicator homogeneously distributed throughout the reaction mixture and capable of generating a second fluorescent signal proportional to
15 the volume of reaction mixture illuminated by the excitation beam;

wherein the lens of the sample interface collects a portion of the first fluorescent signal and the second fluorescent signal and focuses said portion onto the fiber optic, the fiber optic transmitting said portion to a detection and analysis means.

20 2. The system of claim 1 wherein said first fluorescent signal has an intensity and said second fluorescent signal has an intensity, and wherein said detection and analysis means provides a readout comprising a ratio of the intensities of said first fluorescent signal and said second fluorescent signal.

25 3. The system of claim 2 wherein said first fluorescent indicator is a complex-forming dye.

4. The system of claim 3 wherein said system comprises a plurality of said sample interfaces, each said fiber optic of said sample interface having a first end co-axially
30 disposed with said lens and a second end coupled to a fiber optic multiplexer.

5. The system of claim 3 wherein said reaction mixture is contained in a closed reaction vessel, said lens focusing said excitation beam through a portion of a wall of the closed reaction vessel into said reaction mixture, and said portion of the wall being
35 heated so that condensation of components of said reaction mixture thereon is prevented.

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6. The system of claim 3 wherein said system comprises a plurality of said first fluorescent indicators each corresponding to a different said amplification product.

5 7. The system of claim 3 wherein said amplification reaction is a polymerase chain reaction.

8. The system of claim 3 wherein said amplification reaction is a ligase chain reaction

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9. The system of claim 2 wherein said amplification reaction is a polymerase chain reaction and wherein said first fluorescent indicator and said second fluorescent indicator are covalently attached to an oligonucleotide having a nucleotide sequence complementary to a portion of a strand of said amplification product, said second fluorescent indicator quenching the fluorescence of said first fluorescent indicator.

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10. The system of claim 9 wherein said system comprises a plurality of said sample interfaces, each said fiber optic of said sample interface having a first end co-axially disposed with said lens and a second end coupled to a fiber optic multiplexer.

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11. The system of claim 9 wherein said reaction mixture is contained in a closed reaction vessel, said lens focusing said excitation beam through a portion of a wall of the closed reaction vessel into said reaction mixture, and said portion of the wall being heated so that condensation of components of said reaction mixture thereon is prevented.

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12. The system of claim 9 wherein said system comprises a plurality of said first fluorescent indicators each corresponding to a different said amplification product.

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